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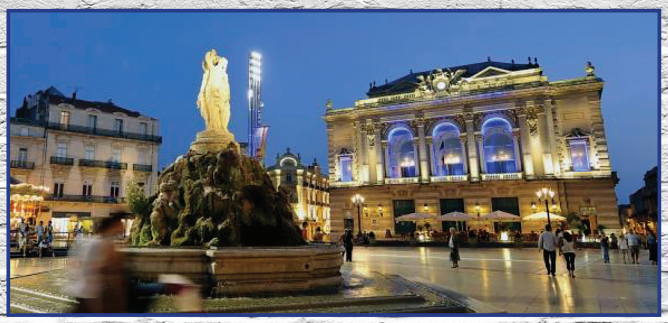

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STUDY OF THE BACTERIAL COMMUNITIES OF THE SEAWEED (*Ulva* spp.) HOLOBIONT TO BASE MANAGEMENT STRATEGIES FOR THE CONTROL OF HARMFUL BACTERIA IN IMTA-RAS

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Bacterial communities associated to *Ulva* spp. (Ulvales, Chlorophyta) are different from those of the surrounding water both in terms of biodiversity and function and play an essential role for the function of the algae, implying that the macroalgae and all their associated microbiota form a singular entity or holobiont (Egan et al 2012). The specific composition of those bacterial communities may be formed by different members of equivalent functional guilds and a stable core set of functional genes in the bacterial communities associated with different *Ulva* species has been demonstrated (Roth-Schulze et al 2018). *Ulva* spp. provide an important niche for biofilm-forming bacteria, including those belonging to the genus *Phaeobacter* with antagonistic activity towards fish pathogens, such as *Vibrio anguillarum* (Prol-García and Pintado 2013). Based on the flexibility in colonization patterns and on the mentioned antagonistic properties of *Phaeobacter*, the possibility of experimental colonization of *Ulva* spp. with antagonistic *Phaeobacter* strains, previously isolated from *Ulva* species, has been demonstrated (Pintado et al 2017) and small-scale trials showed a probiotic effect of *P. gallaeciensis*-colonised *Ulva*, decreasing the mortality of *V. anguillarum*-infected turbot larvae. However, the environmental conditions for *Ulva* spp. culture (agitation and aeration and high light intensity) would have a determinant influence on the maintenance of the biofilms and the production of TDA.

The aim of the research was to study the bacterial communities of different *Ulva* species and the effect of culture and experimental colonization with different species of *Phaeobacter*, studying the influence of factors as light and agitation on bacterial epiphytic communities in *Ulva* spp. and on the colonization by *P. gallaeciensis*.

Samples were taken at different times for bacterial community analysis by PCR-DGGE. Small-scale experiments were conducted with algae thallus discs of 2 cm diameter of different species (*U. austrails*, *U. rigida* and *U. ohnoi*) in well plates with 10 ml sterile Guillard's F/2 medium adjusted to 20 $mg.L^{-1}$ of N (from nitrate) to mimic the concentration on fish-algae IMTA-RAS systems. Algae cultures were inoculated with *P. gallaeciensis* or *P. inhibens* (10^7 CFU ml^{-1}) and controls were conducted in parallel without addition of bacteria. The plates were cultured at 18ºC and 80 rpm orbital agitation, with a daylight-type LED panel and a 12:12 photoperiod. Scale-up of the selected combination *U. ohnoi – Ph. gallaeciensis* was done up to 40L of non-sterile F/2-N medium.

DGGE profiles showed different bacterial communities between *Ulva* species and locations, in samples collected from the sea. Culturing promoted changes on epiphytic bacterial communities, which were affected by the introduction of the *Phaeobacter* strains. Shifts were also observed in the different steps of the scale-up. The study of the effect of light intensity (300, 170, 100 e 50 µmol m^{-2} s^{-1}) and agitation is currently in progress. The results will permit to define the culturing conditions that would favour the establishment of *Phaeobacter* biofilms in *Ulva* spp. and base management strategies for the control of harmful bacteria in IMTA-RAS systems.

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